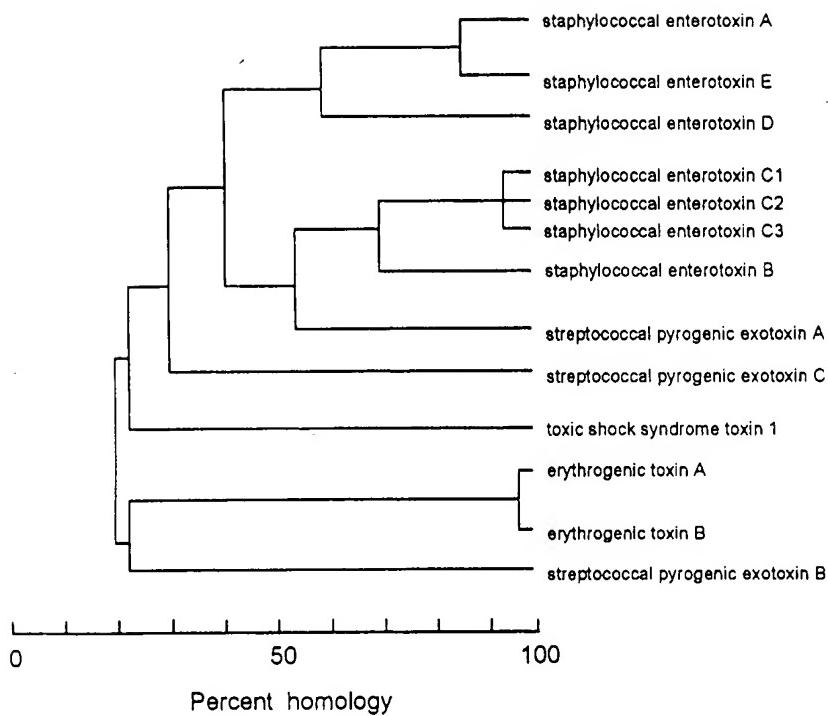


100
90
80
70
60
50
40
30
20
10



This:

FIGURE 1

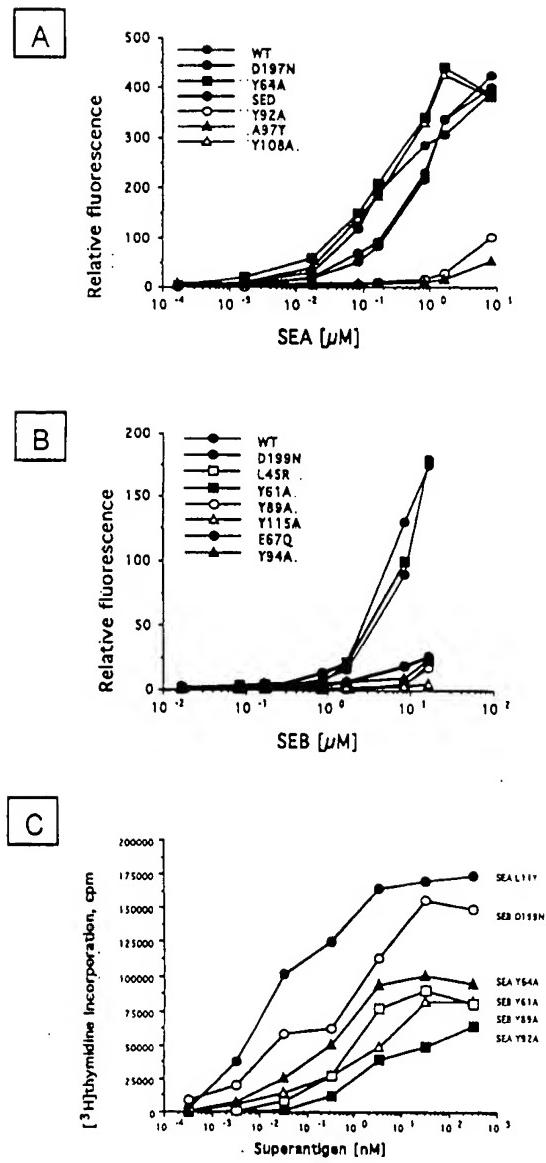
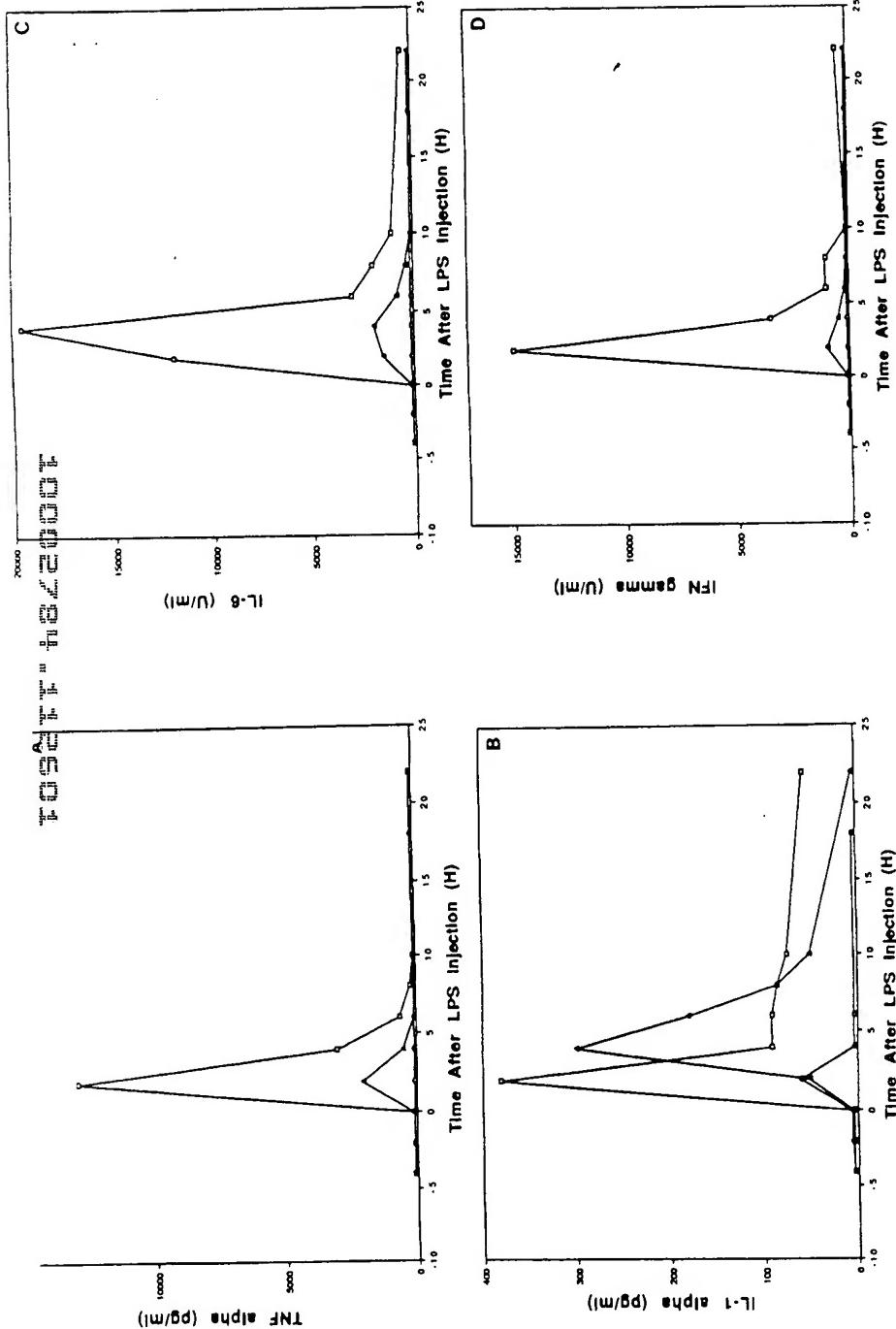
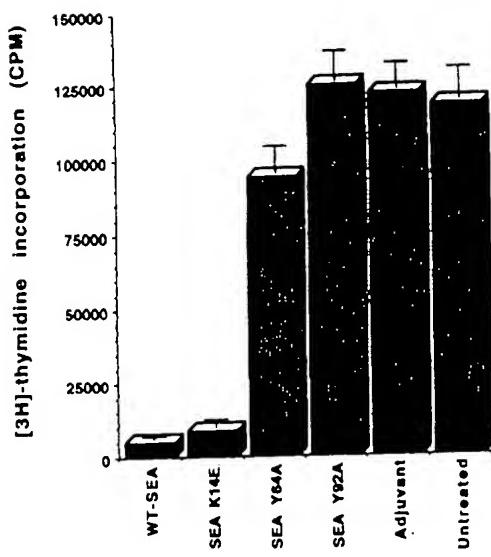


Fig. 2

SEA	... SHDQF QHTILFKGFFTDHSWYMDLLV FDSKDIVDKYK . GKKVDLYGAY GYQCA	SED	... TGDOF ENTLYKKFFTDOLINFDLLI FNSKEMAQHFK . SKNVDVYPIR SINCY	SEE	... SDDQF ENTLLFKGFFTGHPMYNDLLV LGSKDATNKYK . CKKVDLGYAK YYQCYFSKKTNDINSHQTDKRKT . CM	SEB	... SDDQF YFDLJYS IKDTKLGNYDNRV FKNKDADKYK . DKYDVFGAN YYQCYFSKKTNDINSHQTDKRKT . CM	SEC1	... SVDFKF AHDLYNISDKKLKNNYDKVKT LLNEGLAKKYK . DEVDDVYGSN YVNCYFSSSKDNVGKVVTGG . . . KT . CM	SEC2	... SVDFKF AHDLYNISDKKLKNNYDKVKT LLNEGLAKKYK . DEVDDVYGSN YVNCYFSSSKDNVGKVVTGG . . . KT . CM	SEC3	... SVDFKF AHDLYNISDKKLKNNYDKVKT LLNEGLAKKYK . DEVDDVYGSN YVNCYFSSSKDNVGKVVTGG . . . KT . CM	SPEa	... SVDQL SHDLIYNVSG . . . PNYDKLKT LKNQEMATLFK . DKNVDIYGVE YHLCYLCENAE RSACI : GGVTNHEGNHLEIPK	TSST1	... VLDNS1 GSNR1KNTD GSISL1 FPPSPYYSPAFTGEKVDLNTKR KKSQHTSEG TYIHF . Q SGTNT EKLPT . . . P
48	70	92	108										





+15,5

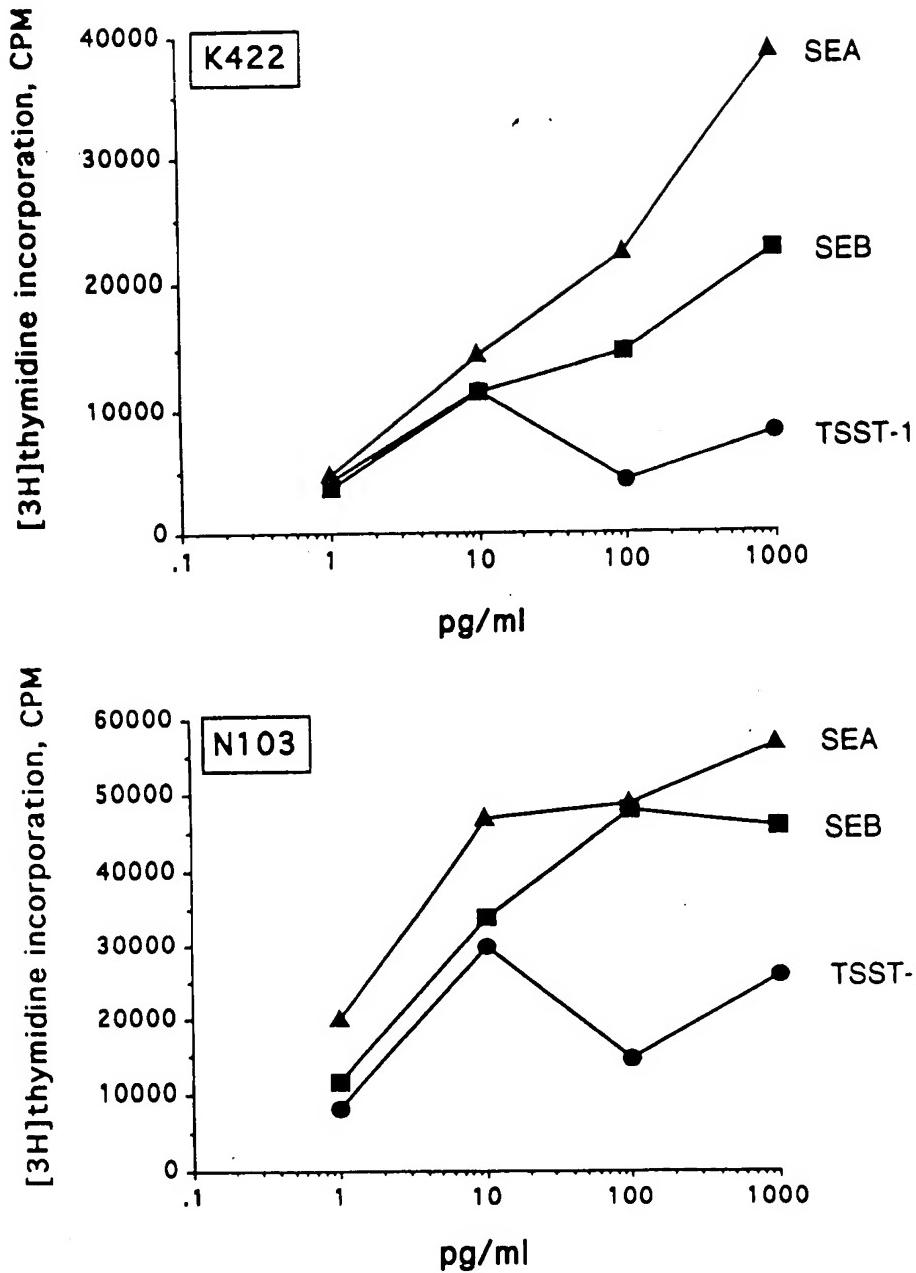


Fig. 6

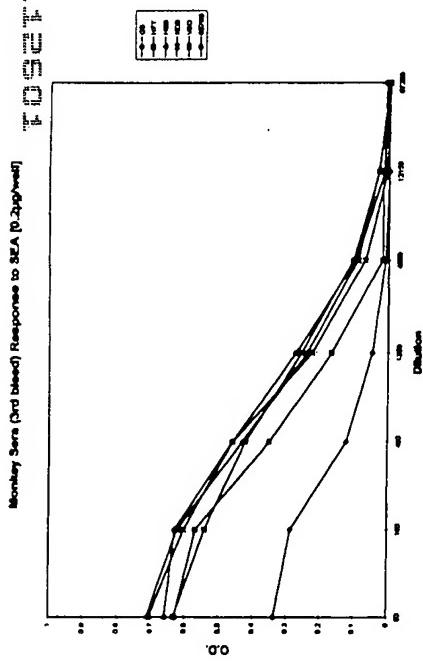
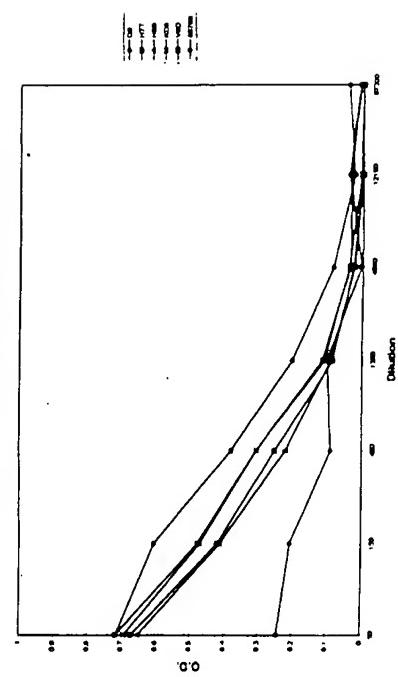
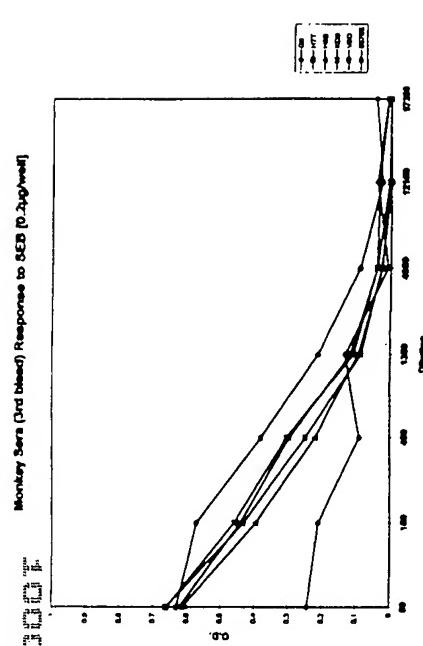
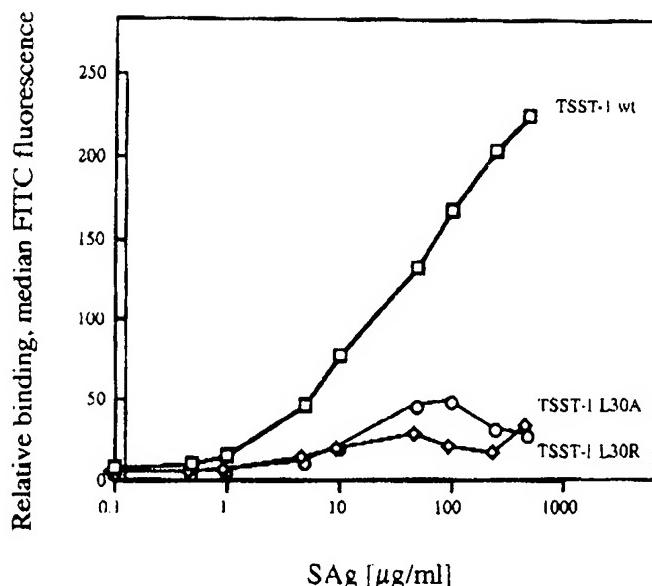


Fig. 7

A.



B.

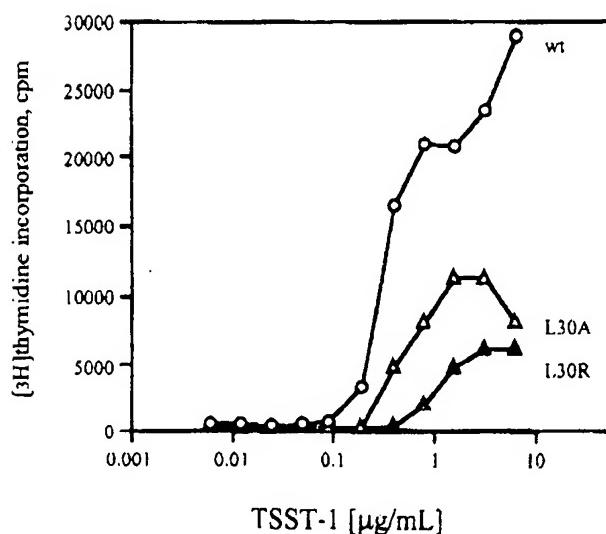
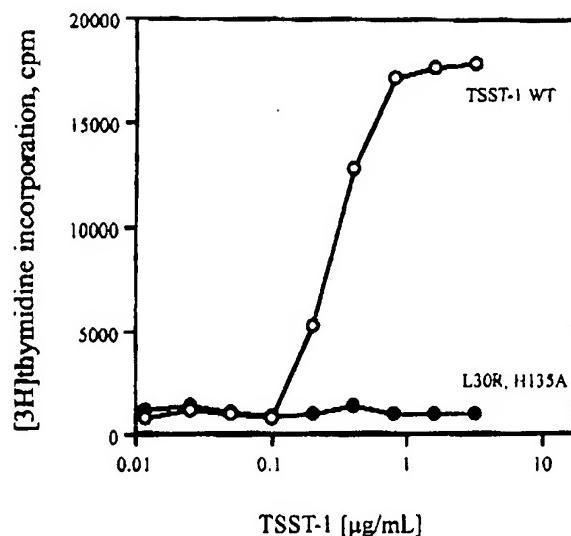


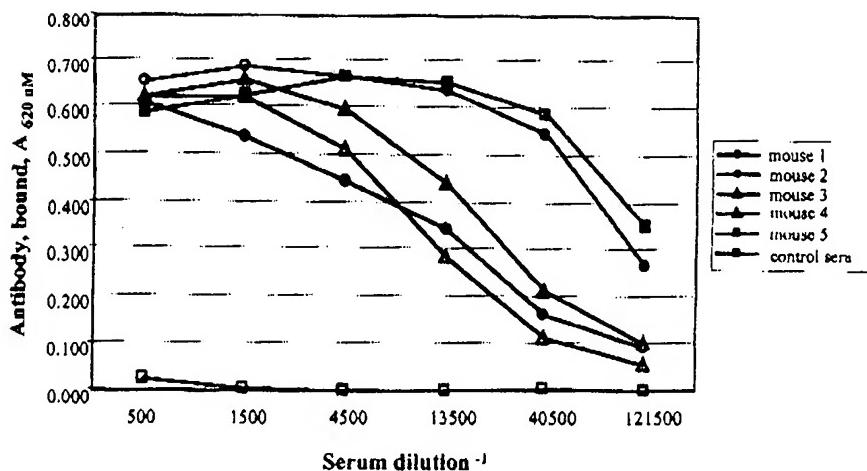
FIGURE 8

C.



Biological activities of TSST-1 mutants. a, Mutations of TSST-1 at amino acid position 30 (L30R, L30A) results in greatly diminished interactions with cell surface HLA-DR, measured by laser fluorescence-activated flow cytometry and FITC-labeled rabbit anti-TSST-1 antibody (affinity purified). b, Mutations of TSST-1 at amino acid position 30 (L30R, L30A) results in greatly diminished activation of human lymphocytes; c, Introduction of an additional mutation, H135A to the TSST-1 mutant L30R results in the maximum reduction in T-cell stimulation. Human T-cell proliferation, was assessed by $[^3\text{H}]$ thymidine incorporation, using a 12 h pulse with label and harvesting cells after 60 h of culture. Each data point represents the mean of triplicate determinations; SEM <5%.

FIGURE 8



Antibody response to TSST-1 mutant L30R. Mice received a total of three injections of vaccine (20 µg/mouse) in Alhydrogel, two weeks between injections. Sera were sampled two weeks after last vaccination and anti-TSST-1 specific antibody was measured by ELISA, using plates coated with wild-type TSST-1. Pooled non-immune mouse sera were used as negative control.

FIGURE 9

A.

F C S E F F " H G A " 2 0 0 0 0

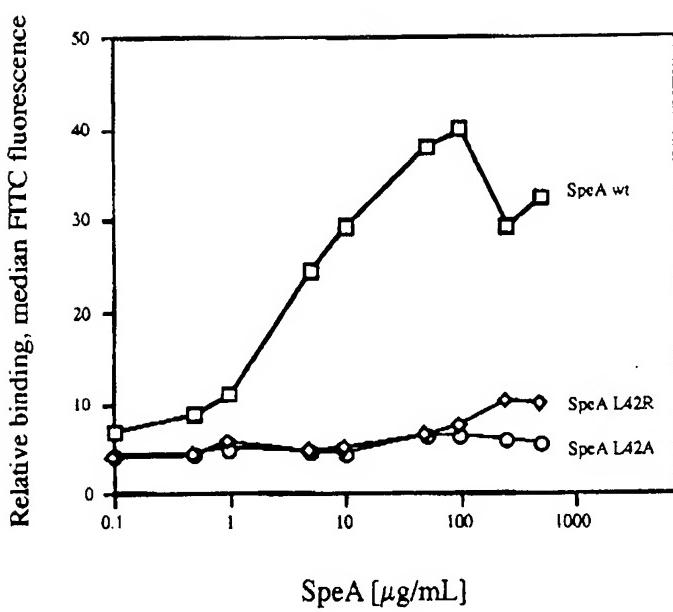
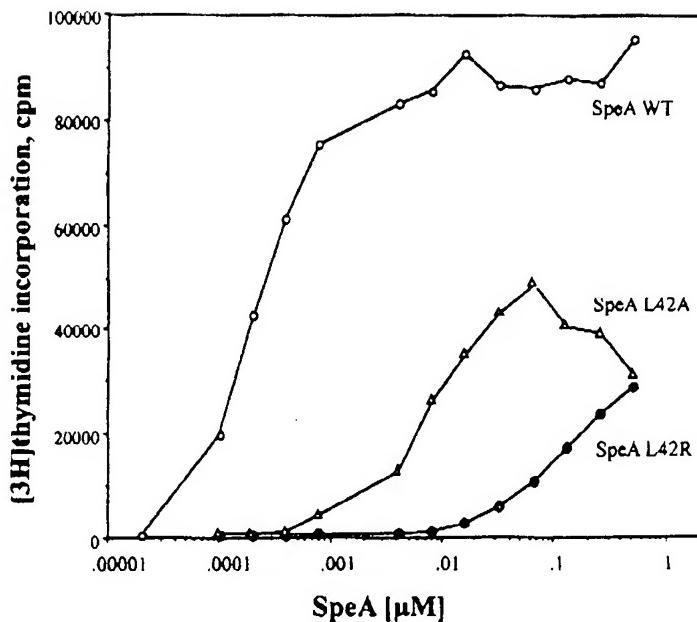


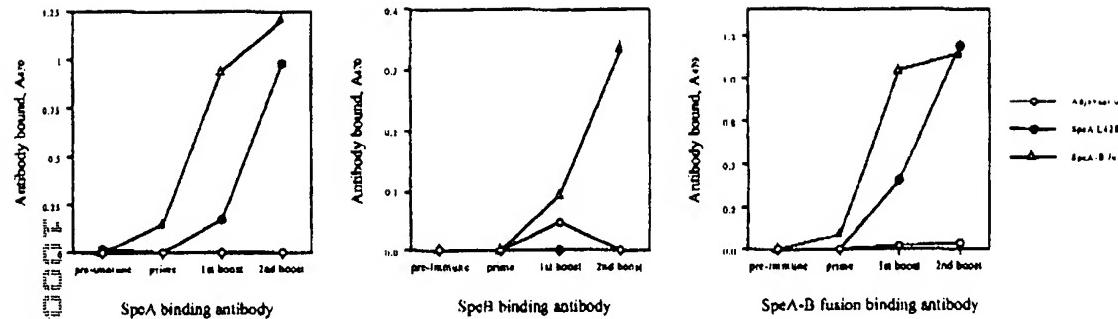
FIGURE 10

B.



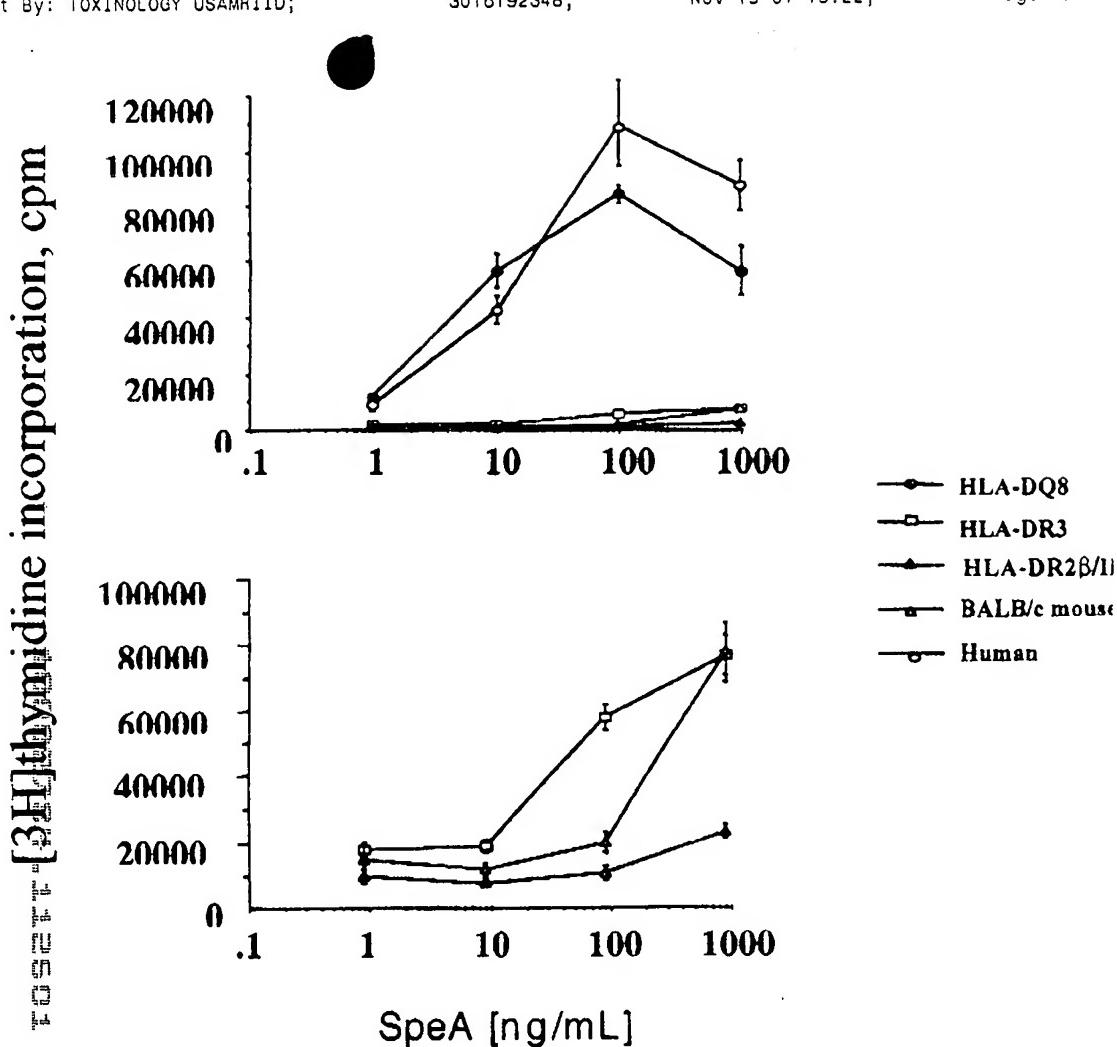
Biological activities of SpeA mutants. a, Mutations of SpeA at amino acid position 42 (L42R) results in greatly diminished interactions with cell surface HLA-DR, measured by laser fluorescence-activated flow cytometry and FITC-labeled rabbit anti-SpeA antibody (affinity purified). b, Mutations of SpeA at amino acid position 42 (L42R or L42A) results in greatly diminished activation of human lymphocytes. Human T-cell proliferation, was assessed by $[^{3\text{H}}]\text{thymidine incorporation}$, using a 12 h pulse with label and harvesting cells after 60 h of culture. Each data point represents the mean of triplicate determinations; SEM <5%.

FIGURE 10



Mouse antibody response to SpeA L42R and SpeA-B fusion constructs. BALB/c mice were vaccinated three times with 10 µg plus adjuvant (MPL™ + TDM+ CWS Emulsion, RJBI ImmunoCHem Research, Inc., Hamilton, MT) of each construct, allowing two weeks between injections. Sera from each experimental group ($n=5$) were pooled for measurement of specific antibodies. Data shown are antigen-specific antibodies (ELISA units) present in a 1:100,000 dilution of pooled sera from mice vaccinated with SpeA L42R, SpeA-B fusion or adjuvant only.

FIGURE 11



T-cell response *in vitro* of mononuclear cells from transgenic mice expressing HLA-DQ8 $\alpha\beta$ and human CD4 closely approximate the physiological response of humans. Mononuclear cells were isolated from spleens of transgenic mice expressing HLA-DR3, HLA-DQ8 or HLA-DR2 β /1E α , or non-transgenic BALB/c mice and human peripheral blood (4 x 10⁵/well). Following 60 h culture with SpeA, cells were pulse-labeled (12 h) with 1 μ Ci of [³H]thymidine. DNA from cells was harvested onto fiberglass filters and incorporated radioactivity measured by liquid scintillation.

FIGURE 12